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(*E*)-1,3-Dialkyl-7-methyl-8-(3,4,5-trimethoxystyryl)xanthines: Potent and Selective Adenosine A_2 Antagonists

Adenosine receptors are localized virtually in all tissues, and modulate a wide range of physiological functions.[1] Adenosine receptors are divided into two major subtypes, designated as A_1 and A_2. The two receptor subtypes can be distinguished by the structure–activity relationships of adenosine agonists and have opposite effects on adenylate cyclase.[2,3]

The methylxanthines theophylline (1, Figure 1) and caffeine (2) exhibit a variety of pharmacological actions primarily through blockade of adenosine receptors.[4] However, they are virtually nonselective antagonists and have weak affinity for A_1 and A_2 receptors. Efforts to develop more potent and highly selective antagonists[5-17] have focused on the modification at the 1-, 3-, 7-, and 8-position of xanthines. Introduction of the propyl group to the 1- and 3-position increases the affinity at A_1 and A_2 receptors.[7-10] The discovery[8,10,15] that cycloalkyl substituents at the 8-position markedly enhanced the affinity at the A_1 receptor have resulted in potent and selective A_1 antagonists such as 8-cyclopentyl-1,3-dipropylxanthine (4)[16] and 1,3-dipropyl-8-(3-noradamantyl)xanthine (5).[17b,c]

(1) Daly, J. W. Adenosine Receptors: Targets for Future Drugs. *J. Med. Chem.* 1982, *25*, 197–207.
(2) Londos, C.; Wolff, J. Two Distinct Adenosine-Sensitive Sites on Adenylate Cyclase. *Proc. Natl. Acad. Sci. U.S.A.* 1977, *74*, 5482–5486.
(3) Van Calker, D.; Müller, M.; Hamprecht, B. Adenosine Regulates via Two Different Types of Receptors, the Accumulation of Cyclic AMP in Cultured Brain Cells. *J. Neurochem.* 1979, *33*, 999–1005.
(4) Fredholm, B. B. Are Methylxanthine Effects due to Antagonism of Endogenous Adenosine? *Trends Pharmacol. Sci.* 1980, *1*, 129–132.
(5) Williams, M. Adenosine Antagonists. *Med. Res. Rev.* 1989, *9*, 219–243.
(6) Bruns, R. F. Structure Activity Relationships for Adenosine Antagonists. In *Purines in Cellular Signaling: Targets for New Drugs*; Jacobson, K. A., Daly, J. W., Manganiello, V., Eds.; Springler: New York, 1990; pp 126–135.
(7) Bruns, R. F.; Daly, J. W.; Snyder, S. H. Adenosine Receptor Binding: Structure–Activity Analysis Generates Extremely Potent Xanthine Antagonists. *Proc. Natl. Acad. Sci. U.S.A.* 1983, *80*, 2077–2080.
(8) Bruns, R. F.; Lu, G. H.; Pugsley, T. A. Characterization of the A_2 Adenosine Receptor Labeled by [^{3}H]NECA in Rat Striatal Membranes. *Mol. Pharmacol.* 1986, *29*, 331–346.
(9) Schwabe, U.; Ukena, D.; Lohse, M. J. Xanthine Derivatives as Antagonists at A_1 and A_2 Adenosine Receptors. *Naunyn-Schmiedeberg's Arch. Pharmacol.* 1985, *330*, 212–221.
(10) Martinson, E. A.; Johnson, R. A.; Wells, J. N. Potent Adenosine Receptor Antagonists that are Selective for the A_1 Receptor Subtype. *Mol. Pharmacol.* 1987, *31*, 247–252.
(11) (a) Hamilton, H. W.; Ortwine, D. F.; Worth, D. F.; Badger, E. W.; Bristol, J. A.; Bruns, R. F.; Haleen, S. J.; Steffen, R. P. Synthesis of Xanthines as Adenosine Antagonists, a Practical Quantiative Structure–Activity Relationship Application. *J. Med. Chem.* 1985, *28*, 1071–1079. (b) Daly, J. W.; Padgett, W.; Shamim, M. T.; Butts-Lamb, P.; Waters, J. 1,3-Dialkyl-8-(*p*-sulfophenyl)xanthines: Potent Water-Soluble Antagonists for A_1- and A_2-Adenosine Receptors. *J. Med. Chem.* 1985, *28*, 487–492. (c) Daly, J. W.; Padgett, W. L.; Shamim, M. T. Analogues of 1,3-Dipropyl-8-phenylxanthines: Enhancement of Selectivity at A_1-Adenosine Receptors by Aryl Substituents. *J. Med. Chem.* 1986, *29*, 1520–1524.
(12) (a) Jacobson, K. A.; Kirk, K. L.; Padgett, W. L.; Daly, J. W. Functionalized Congeners of 1,3-Dialkylxanthines: Preparation of Analogues with High Affinity for Adenosine Receptors. *J. Med. Chem.* 1985, *28*, 1334–1340. (b) Jacobson, K. A.; Kirk, K. L.; Padgett, W. L.; Daly, J. W. A. Functionalized Congener Approach to Adenosine Receptor Antagonists: Amino Acid Conjugates of 1,3-Dipropylxanthine. *Mol. Pharmacol.* 1986, *29*, 126–133. (c) Jacobson, K. A.; Ukena, D.; Padgett, W.; Daly, J. W.; Kirk, K. L. Xanthine Functionalized Congeners as Potent Ligands at A_2-Adenosine Receptors. *J. Med. Chem.* 1987, *30*, 211–214.
(13) (a) Daly, J. W.; Padgett, W. L.; Shamim, M. T. Analogues of Caffeine and Theophylline: Effect of Structural Alterations on Affinity at Adenosine Receptors. *J. Med. Chem.* 1986, *29*, 1305–1308. (b) Ukena, D.; Shamim, M. T.; Padgett, W.; Daly, J. W. Analogs of Caffeine: Antagonists with Selectivity for A_2 Adenosine Receptors. *Life Sci.* 1986, *39*, 743–750. (c) Shamim, M. T.; Ukena, D.; Padgett, W. L.; Daly, J. W. Effects of 8-Phenyl and 8-Cycloalkyl Substituents on the Activity of Mono-, Di-, and Trisubstituted Alkylxanthines with Substitution at the 1-, 3-, and 7-Positions. *J. Med. Chem.* 1989, *32*, 1231–1237. (d) Daly, J. W.; Hide, I.; Müller, C. E.; Shamim, M. Caffeine Analogs: Structure–Activity Relationships at Adenosine Receptors. *Pharmacology* 1991, *42*, 309–321.
(14) Bruns, R. F.; Fergus, J. H.; Badger, E. W.; Bristol, J. A.; Santay, L. A.; Hays, S. J. PD 115,199: An Antagonist Ligand for Adenosine A_2 Receptors. *Naunyn-Schmiedeberg's Arch. Pharmacol.* 1987, *335*, 64–69.
(15) (a) Shamim, M. T.; Ukena, D.; Padgett, W. L.; Daly, J. W. 8-Aryl- and 8-Cycloalkyl-1,3-dipropylxanthines: Further Potent and Selective Antagonists for A_1-Adenosine Receptors. *J. Med. Chem.* 1988, *31*, 613–617. (b) Katsushima, T.; Nieves, L.; Wells, J. N. Structure–Activity Relationships of 8-Cycloalkyl-1,3-dipropylxanthines as Antagonists of Adenosine Receptors. *J. Med. Chem.* 1990, *33*, 1906–1910. (c) Jacobson, K. A.; Kiriasis, L.; Barone, S.; Bradbury, B. J.; Kammula, U.; Campagne, J. M.; Secunda, S.; Daly, J. W.; Neumeyer, J. L.; Pfleiderer, W. Sulfur-Containing 1,3-Dialkyl xanthine Derivatives as Selective Antagonists at A_1-Adenosine Receptors. *J. Med. Chem.* 1989, *32*, 1873–1879. (d) Erickson, R. H.; Hiner, R. N.; Feeney, S. W.; Blake, P. R.; Rzeszotarski, W. J.; Hicks, R. P.; Costello, D. G.; Abreu, M. E. 1,3,8-Trisubstituted Xanthines. Effects of Substitution Pattern upon Adenosine Receptor A_1/A_2 Affinity. *J. Med. Chem.* 1991, *34*, 1431–1435.
(16) (a) Bruns, R. F.; Fergus, J. H.; Badger, E. W.; Bristol, J. A.; Santay, L. A.; Hartman, J. D.; Hays, S. J.; Huang, C. C. Binding of the A_1-Selective Adenosine Antagonist 8-Cyclopentyl-1,3-dipropylxanthine to Rat Brain Membranes. *Naunyn-Schmiedeberg's Arch. Pharmacol.* 1987, *335*, 59–63. (b) Lohse, M. J.; Klotz, K.-N.; Lindenborn-Fotinos, J.; Reddington, M.; Schwabe, U.; Olsson, R. A. 8-Cyclopentyl-1,3-dipropylxanthine (DPCPX)—A Selective High Affinity Antagonist Radioligand for A_1 Adenosine Receptors. *Naunyn-Schmiedeberg's Arch. Pharmacol.* 1987, *336*, 204–210.
(17) (a) Shimada, J.; Suzuki, F.; Nonaka, H.; Karasawa, A.; Mizumoto, H.; Ohno, T.; Kubo, K.; Ishii, A. 8-(Dicyclopropylmethyl)-1,3-dipropylxanthine: A Potent and Selective Adenosine A_1 Antagonist with Renal Protective and Diuretic Activities. *J. Med. Chem.* 1991, *34*, 466–469. (b) Suzuki, F.; Shimada, J.; Ishii, A.; Ohno, T.; Karasawa, A.; Kubo, K.; Nonaka, H. Xanthine Compounds. Eur. Patent 415 456, 1990. (c) Shimada, J.; Suzuki, F.; Nonaka, H.; Ishii, A. 8-Polycycloalkyl-1,3-dipropylxanthines as Potent and Selective Antagonists for A_1-Adenosine Receptors. *J. Med. Chem.* 1992, *35*, 924–930.

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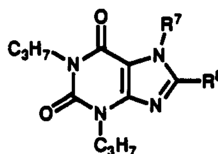
Table I. A_1 and A_2 Adenosine Receptor Binding of 8-Substituted-1,3-dipropylxanthines



no.	R^7	R^8	K_i[a] nM A_1	A_2	K_i ratio A_1/A_2
8	H	2-phenylethyl	560 (57.8)[b]	6200 (593)[c]	0.090
9	methyl	2-phenylethyl	1300	2200	0.59
10	H	(*E*)-styryl	1800 ± 750	26 ± 4.5	69
11	methyl	(*E*)-styryl	720 ± 340	15 ± 5.9	48
12	H	(*E*)-α-methylstyryl	>100000	>10000	
13	methyl	(*E*)-α-methylstyryl	>10000	>10000	
14	H	(*E*)-cinnamyl	870	1600	0.54
15	methyl	(*E*)-cinnamyl	3500	1800	1.9
16	H	2-cyclopentylethyl	320	6000	0.053
17	methyl	2-cyclopentylethyl	1300	>10000	
18[d]	methyl	cyclopentyl	8100 ± 2200 (2300)[e]	>100000 (220)[f]	0.26
1		(theophylline)	23000 ± 330 (13000)[e] (8470)[g]	16000 ± 2200 (14000)[f] (25300)[h]	1.4
2		(caffeine)	100000 ± 2000 (44000)[e] (29100)[g]	27000 ± 1700 (30000)[f] (48100)[h]	3.7
3		(1,3-dipropylxanthine)	1200 ± 120 (450)[g]	2400 ± 420 (5160)[h]	0.5
4		(8-cyclopentyl-1,3-dipropylxanthine)	6.4 ± 0.35 (0.23)[b] (0.9)[e] (0.46)[g]	590 ± 48 (230)[c] (140)[f] (410)[h]	0.011
5		(1,3-dipropyl-8-(3-noradamantyl)xanthine)	1.3 ± 0.12	380 ± 30	0.0034
6		(PD-115199)	140 (13.9)[i]	26 (15.5)[h]	5.4
7		(XAC)	11 (1.2)[j]	21 (63)[h]	0.52

[a] A_1 binding was carried out with N^6-[^{3}H]cyclohexyladenosine in guinea pig forebrain membranes as described,[18] and A_2 binding was carried out with *N*-[^{3}H]ethyladenosin-5′-uronamide in the presence of 50 nM cyclopentyladenosine in rat striatal membranes.[8] Concentration–inhibition curves were carried out in duplicate with five or more concentrations of each test agent, and IC_{50} values were calculated from computerization of logit log curve. IC_{50} values were converted to K_i values as described.[20] When the assays were carried out three or more times, standard errors (SEM) are given in the table. Xanthines were dissolved in aqueous dimethyl sulfoxide and the final concentration of dimethyl sulfoxide in the assay was less than 0.9%.[17c] [b] A_1 binding measured as inhibition of N^6-[^{3}H]cyclohexyladenosine to rat cortical membranes.[15d] [c] A_2 binding measured as inhibition of *N*-[^{3}H]ethyladenosin-5′-uronamide to rat striatal membranes.[15d] [d] H: calcd, 8.23; found, 8.87. [e] A_1 binding measured as inhibition of (*R*)-N^6-([^{3}H]phenylisopropyl)adenosine to rat cortical membranes.[13c] [f] K_B values for inhibition of adenylate cyclase stimulation by *N*-[^{3}H]ethyladenosin-5′-uronamide in human platelet membranes.[13c] [g] A_1 binding measured as inhbition of [^{3}H]-N^6-cyclohexyladenosine to whole brain membranes.[8,16a] [h] A_2 binding measured as inhibition of *N*-[^{3}H]ethyladenosin-5′-uronamide to rat striatal membranes.[8,14,15c] [i] A_1 binding measured as inhibition of [^{3}H]-N^6-cyclohexyladenosine to rat cortical membranes.[14] [j] A_1 binding measured as inhibition of [^{3}H]-(*R*)-N^6-(phenylisopropyl)adenosine to rat cortical membranes.[15c]

Although selective A_1 antagonists have been found, no antagonist with high selectivity toward the A_2 receptor has been forthcoming. Some caffeine derivatives[13] such as 3,7-dimethyl-1-propargylxanthine or 1,3-dipropyl-7-methylxanthine have been reported to possess a moderate degree of A_2 selectivity. Surprisingly, 8-cycloalkyl substituents (cyclopentyl and cyclohexyl) increase the affinity of caffeine and 1,3-dipropyl-7-methylxanthine at the A_2 receptor.[13c] Introduction of some para-substituted phenyl groups such as 4-[[2-(dimethylamino)ethyl]methylsulfamoyl]phenyl (**6**; PD-115199)[11a,14] or 4-[[(2-aminoethyl)amino]carbonyl]methoxy]phenyl (**7**; XAC)[12] into the 8-position potently enhanced the affinity at A_1 and A_2 receptors. This observation suggests that a different pocket from that recognized by 8-cycloalkyl substituents exists in A_1 and A_2 receptors. The present study describes a potent and selective adenosine A_2 antagonist, a series of (*E*)-1,3-dialkyl-7-methyl-8-styrylxanthine derivatives which contains a new hydrophobic moiety at the 8-position.

The potency of the xanthine derivatives at adenosine A_1 and A_2 receptors was determined by standard radioligand binding procedures. Adenosine A_1 binding was performed with N^6-[^{3}H]cyclohexyladenosine binding in guinea pig forebrain membranes[18] which is the most similar to that in man.[19] A_2 receptor binding was performed with *N*-[^{3}H]ethyladenosin-5′-uronamide ([^{3}H]NECA) in rat

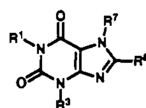
(18) Bruns, R. F.; Daly, J. W.; Snyder, S. H. Adenosine Receptors in Brain Membranes: Binding of N^6-Cyclohexyl[^{3}H]adenosine and 1,3-Diethyl-8-[^{3}H]phenylxanthine. *Proc. Natl. Acad. Sci. U.S.A.* **1980**, *77*, 5547–5551.

(19) (a) Ferkany, J. W.; Valentine, H. L.; Stone, G. A.; Williams, M. Adenosine A_1 Receptors in Mammalian Brain: Species Differences in Their Interactions with Agonists and Antagonists. *Drug. Dev. Res.* **1986**, *9*, 85–93. (b) Ukena, D.; Jacobson, K. A.; Padgett, W. L.; Ayala, C.; Shamim, M. T.; Kirk, K. L.; Olsson, R. A.; Daly, J. W. Species Differences in Structure–Activity Relationships of Adenosine Agonists and Xanthine Antagonists at Brain A_1 Adenosine Receptors. *FEBS Lett.* **1986**, *209*, 122–128. (c) Stone, G. A.; Jarvis, M. F.; Sills, M. A.; Weeks, B.; Snowhill, E. W.; Williams, M. Species Differences in High-Affinity Adenosine A_2 Binding Sites in Striatal Membranes from Mammalian Brain. *Drug Dev. Res.* **1988**, *15*, 31–46.

Table II. A_1 and A_2 Adenosine Receptor Binding of (*E*)-8-Styryl-1,3-dipropylxanthines

no.	R^7	Ar	K_i,[a] nM A_1	K_i,[a] nM A_2	K_i ratio A_1/A_2
10	H	phenyl	1800 ± 750 (22.2)[b]	26 ± 4.5 (85.1)[b]	69
11	methyl	phenyl	720 ± 340	15 ± 5.9	15
19	H	4-methoxyphenyl	>100000	110	
20	methyl	4-methoxyphenyl	1400 ± 860	18 ± 6.3	78
21	H	3,4-dimethoxyphenyl	1700	6700	0.25
22	methyl	3,4-dimethoxyphenyl	1500 ± 780	7.8 ± 2.7	190
23	H	3,4,5-trimethoxyphenyl	850 ± 420	17 ± 1.0	50
24	methyl	3,4,5-trimethoxyphenyl	2100 ± 800	14 ± 2.6	150
25	H	4-chlorophenyl	>100000	>100000	
26	methyl	4-chlorophenyl	>10000	49	
27	H	3,4-dichlorophenyl	>100000	>100000	
28	methyl	3,4-dichlorophenyl	>100000	7500	

[a] See footnote *a* in Table I. [b] See footnotes *b* and *c* in Table I.



no.	R^1	R^3	R^7	R^8
1	methyl	methyl	H	H
2	methyl	methyl	methyl	H
3	propyl	propyl	H	H
4	propyl	propyl	H	cyclopentyl
5	propyl	propyl	H	3-noradamantyl
6	propyl	propyl	H	$SO_2N(CH_3)CH_2CH_2N(CH_3)_2$
7	propyl	propyl	H	$OCH_2CONHCH_2CH_2NH_2$

Figure 1. Chemical structures of reference compounds.

striatal membranes.[8] Table I shows a series of 1,3-dipropylxanthines containing various hydrophobic substituents at the 8-position with K_i values. (*E*)-Styryl substitution (**10**) had about 100-fold higher affinity at the A_2 receptor than a parent compound (**3**) and resulted in high A_2 selectivity (69-fold). 2-Phenylethyl (**8**) or (*E*)-cinnamyl (**14**) substitution did not cause such enhancement of affinity at the A_2 receptor. Incorporation of methyl group into the vinylene group (**12**) caused reduction of affinity at A_1 and A_2 receptors. Therefore the vinylene group between the xanthine and the phenyl group seemed to play an important role for the receptor interactions.

7-Methyl substitution did not alter the affinity at A_1 and A_2 receptors in 8-(2-phenylethyl)-, (*E*)-styryl-, and (*E*)-cinnamylxanthines (compare **9**, **11**, **13**, and **15** with **8**, **10**, **12**, and **14**). In contrast to this observation, introduction of methyl group into the 7-position of 8-(2-cyclopentylethyl)- or 8-cyclopentyl-substituted xanthine resulted in the decreased affinity at the A_2 receptor (compare **17** and **18** with **16** and **4**). Consequently, the electrostatic effects of the styryl or cinnamyl group appeared to be more favorable for their interactions with the A_2 receptor than those of the cyclopentyl group.

The activity of compound **18** at the A_2 receptor was lower than the reported activity in Shamim's work.[13c] This discrepancy seems to be arisen from the different assay system. They used inhibitory activity of NECA-elicited stimulation of adenylate cyclase (human platelet membranes) as evaluation of affinity at the A_2 receptor.

Since (*E*)-8-styrylxanthines (**10** and **11**) were selective and potent A_2 antagonists, the effects of substituents in the styryl phenyl group on affinity at the A_2 receptor were examined (Table II). Introduction of chloro or methoxy substituents into the phenyl group of (*E*)-1,3-dipropyl-8-styrylxanthine (**10**) resulted in the decreased activity to A_1 and A_2 receptors (compare **19**, **21**, **25**, and **27** with **10**) except for a (*E*)-3,4,5-trimethoxystyryl derivative (**23**). On the other hand, introduction of two or three methoxy groups into the styryl phenyl group of (*E*)-1,3-dipropyl-7-methyl-8-styrylxanthine (**11**) enhanced the A_2 selectivity (compare **22** and **24** and **11**). In contrast to the result in Table I, 7-methyl substitution in these derivatives increased the A_2 selectivity (compare **19**, **21**, **25**, and **27** with **20**, **22**, **26**, and **28**). Compound **10** showed a big species difference in A_1 receptor binding (rat cortical membrane, K_i = 22.2 nM;[15d] guinea pig forebrain membrane, K_i = 1800 nM). Thus the A_1 binding of several compounds was carried out with [^{3}H]CHA using rat forebrain membranes as described before.[7,16a] K_i values of compound **10**, **11**, **20**, **22**, and **24** were 35 ± 1.0, 220 ± 78, 340 ± 46, 430 ± 150, and 1100 ± 380 nM, respectively. These compounds are about 2–4-fold more potent at the A_1 receptor in rat brain than in guinea pig brain except **10**. We need more studies in order to explain an exceptionally big species difference of **10** in A_1 receptor binding.

Since (*E*)-3,4,5-trimethoxystyryl substitution at the 8-position appeared to enhance the affinity at the A_2 receptor in general, the effects of other substituent at the 1- and 3-position were examined (Table III). Compound **31** was less active than compound **23**. Thus alkyl substitution at the 1-position was important for affinity at the A_2 receptor. Introduction of the methyl group into the 7-position of (*E*)-8-(3,4,5-trimethoxystyryl)xanthines enhances the A_2 selectivity in general (compare **29**, **23**, **32**, and **34** with **30**, **24**, **33**, and **35**). Methyl or allyl substitution at the 1- and 3-position was less active at the A_1 receptor (**29**, **30**, **34**, and **35**). No apparent differences in the affinity at the A_2 receptor were observed among these 1,3-disubstituted-7-methylxanthine derivatives (**30**, **24**, **33**, and **35**). This result is greatly contrasting with that of 1,3-disubstituted 8-alkyl-

Table III. A_1 and A_2 Adenosine Receptor Binding of (*E*)-8-(3,4,5-Trimethoxystyryl)xanthines

no.	R^1	R^3	R^7	K_i,[a] nM A_1	A_2	K_i ratio A_1/A_2
29[b]	methyl	methyl	H	>100000	71 ± 8.2	>1100
30[b]	methyl	methyl	methyl	>100000	18 ± 4.2	>5600
23	propyl	propyl	H	850 ± 420	17 ± 1.0	50
24	propyl	propyl	methyl	2100 ± 800	14 ± 2.6	150
31	H	propyl	H	820	2200	0.37
32	butyl	butyl	H	1400	93	15
33	butyl	butyl	methyl	2300	52	44
34	allyl	allyl	H	>100000	47	>2100
35	allyl	allyl	methyl	>100000	15 ± 8.6	>6700

[a] See footnote *a* in Table I. [b] Prepared by published procedures.[22]

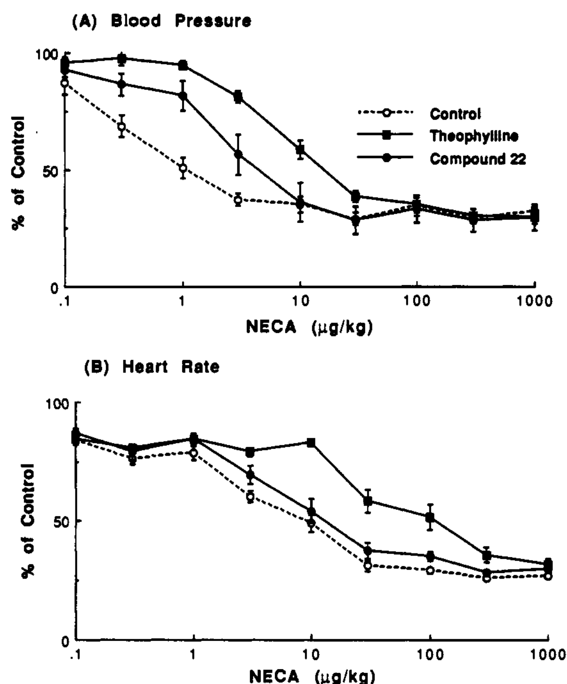


Figure 2. Effect of compound **22** on NECA-induced (A) hypotensive and (B) bradycardic responses in anesthetized rats. The dotted line shows the effects of NECA. Compound **22** or theophylline was suspended with 0.3% Tween 80 and administered orally at the dose of 30 mg/kg. One hour later, increasing doses of NECA were given intravenously and the changes in diastolic blood pressure and heart rate were recorded. Data are expressed as the mean ± SEM (n = 6–10).

or 8-polycycloalkylxanthine derivatives in A_1 receptor binding where 1,3-disubstituents dramatically influenced affinity at the A_1 receptor and its selectivity as previously described.[17]

We then examined the biological activity of the most potent A_2 antagonist **22** in vivo. As shown in Figure 2, NECA caused a dose-dependent decrease in heart rate and in blood pressure in the anesthetized rats.[23] Water solubility of **22** is unfortunately very poor (<10 μg/mL) but ethanol dissolves it to some extent (0.7 mg/mL). Thus **22** was orally administered in 0.3% Tween suspension. Compound **22** produced a much larger rightward shift of the NECA dose–response curve for blood pressure than for heart rate at the dose of 30 mg/kg. By contrast, theophylline, a nonselective antagonist, produced equivalent rightward shifts in the two dose–response curves. Adenosine is supposed to reduce heart rate via an effect on the A_1 receptor and blood pressure via the A_2 receptor.[23] Thus **22** was also identified to be a selective adenosine A_2 antagonist in vivo.

In conclusion, introduction of the (*E*)-3,4-dimethoxystyryl or (*E*)-3,4,5-trimethoxystyryl group into the 8-position of 1,3-dialkyl-7-methylxanthines enhanced the A_2 antagonism.[21] The pharmacological activity of these A_2 antagonists will be reported in due course.

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Supplementary Material Available: Experimental and characterization data for the compounds discussed in this work (6 pages). Ordering information is given on any current masthead page.

(20) Cheng, Y. C.; Prusoff, W. H. Relationship between the Inhibition Constant (K_I) and the Concentration of Inhibitor which Causes 50% Inhibition (I_{50}) of an Enzymatic Reaction. *Biochem. Pharmacol.* **1973**, *22*, 3099–3108.

(21) Suzuki, F.; Shimada, J.; Ishii, A.; Nonaka, H.; Kosaka, N.; Ichikawa, S. Xanthine Derivatives. PCT Patent WO, 92/06976, 1992 (Priority Data: October 18, 1990, Japan Patent Application 280 171).

(22) Schweiss, D.; Long, L. M. 1,3-Dialkyl-7-methyl-8-styrylxanthines as Cerebral Stimulants. Ger. Patent 2 037 171, 1971; *Chem. Abstr.* 1971, *74*, 100108n.

(23) Fredholm, B. B.; Jacobson, K. A.; Jonzon, B.; Kirk, K. L.; Li, Y. O.; Daly, J. W. Evidence That a Novel 8-Phenyl-Substituted Xanthine Derivative is a Cardioselective Adenosine Receptor Antagonist In Vivo. *J. Cardiovasc. Pharmacol.* **1987**, *9*, 396–400.

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